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# Macroinvertebrates and Fishes as Bioindicators of Stream Water Pollution

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## Abstract

Freshwater ecosystems worldwide have been progressively deteriorated during the past decades due to an increasing human pressure that has led to a decrease in aquatic biodiversity. Among the human activities of high impact on freshwater ecosystems is the land-use change, principally from native forests to agriculture. To evaluate the impacts of human activities on water quality, a traditional approach has considered the use of single physical-chemical parameters. However, this approach may be insufficient to fully assess the impact of these human activities on freshwaters. Therefore, there is a need for alternative tools such as the indices of biotic integrity that may provide a complement to traditional approaches. In the literature, there are several examples of biotic indicators that have shown promising results in evaluating water quality including the use of macroinvertebrates and fish diets. Here, we provide a review of the indicators of biotic integrity that included fish assemblages as well as macroinvertebrates as bioindicators. We identify pros and cons of using aquatic communities as indicators of water quality. Finally, we develop a procedure that combines fish and macroinvertebrate assemblages as bioindicators and discuss their effectiveness using illustrative examples from streams under several agricultural uses in the Mediterranean region of Chile.

**Keywords:** biological monitoring, biotic index, macroinvertebrates, fishes, Mediterranean, Chile

## 1. Introduction

Of all the water on earth, freshwater accounts for just 0.01% and covers only 0.8% of the planet's surface [1]. Freshwaters are among the most threatened ecosystems of the world, and thus, understanding their health statuses is of special relevance. Indeed, the physical, chemical, and biological integrities of water are highly important for successfully implementing conservation and management strategies before ecosystem health or biotic integrity are affected [2–4]. This chapter provides a review of known biotic integrity indicators, including benthic macroinvertebrate and fish communities that have been proposed to serve as water quality indicators. In addition, the pros and cons of using aquatic communities as water quality indicators are discussed. Finally, we present a research case study in which benthic macroinvertebrate and fish communities are used as bioindicators, in addition to discussing the effectiveness of using illustrative examples for streams subject to several agriculture uses in a region of Chile dominated by agricultural activities.

Worldwide, a primary threat to freshwater ecosystems is the rapid changes occurring in land uses (**Figure 1**), a situation that has intensified over the past decade [5, 6]. Most recent land use conversion has been for crop production, which notably impacts proximal ecosystems due to changes over extensive crop areas [7]. In particular, the fertilizers and pesticides used in agriculture negatively affect freshwater ecosystems by draining into rivers, where eutrophication and other negative effects, such as high sediment deposits and postsedimentation, subsequently occur. Furthermore, the extensive land use of farming many times results in landscape deforestation, which often arrives to the riverbank itself. This deforestation can increase the temperature of and quantity of light in river water. When coupled with eutrophication, the trophic changes within the aquatic ecosystem can be disturbed, causing, for example, a decreased quantity of aquatic taxa as compared to rivers with fewer alterations [8, 9].



**Figure 1.** Examples of land use in the central-south of Chile. Left: Stream nearby corn crops, right: Stream borderer by native forest of the Maule Region watershed (photographs by P. Fierro).

## 2. Indicators of aquatic ecosystem health

The definition of a healthy ecosystem has been widely debated in the literature. Nevertheless, the definition proposed by Rapport is one of the most widely accepted [10]. This definition states that a healthy ecosystem is defined by the “absence of danger signals in the ecosystem, the ability of the ecosystem to quickly and completely recover (resilience), and/or the lack of risks or threats that push the ecosystem composition, structure, and/or function.” The purpose of monitoring aquatic ecosystem health is to identify physicochemical and biological changes arising from anthropogenic impacts [11]. This information is crucial for managers and policy makers to make informed decisions towards improving the environment and, consequently, human health [12].

Traditional techniques for measuring water quality and to establish aquatic health assess a number of physical and chemical parameters of the water. However, these measurements do not accurately account for the real impacts that physicochemical activities have on freshwater ecosystems [13]. Indeed, these parameters interact and evidence accumulative effects over time, the impacts of which can finally affect aquatic biota [14]. Due to this, other measurements that consider non-natural disturbing effects on ecological integrity should be used to calculate the quality of aquatic resources [15]. Indices based on aquatic biota have been widely successful in determining the integrity of aquatic ecosystems [16].

The use of indices that evaluate water quality through biological parameters, such as freshwater ecosystem structure and performance, has considerably increased in recent years and has gained recognition as an important measure for calculating the global integrity of freshwater ecosystems [17–19]. Biological monitoring is advantageous in that it can integrate and reflect accumulative changes over time, which is in contrast to a number of other methods, such as flow regimen, energetic resources, and biotic interactions [20, 21]. Another benefit is that the high fauna diversity found in aquatic ecosystems, which include microorganisms, algae, periphyton, phytoplankton, zooplankton, macroinvertebrates, fish, and mammals, can be included in evaluations of river health [4].

Among fauna, fish and macroinvertebrate assemblages have been highlighted as good bioindicators for monitoring ecosystem degradation related to farming and forestry, as well as to urban and industrial effluents [9, 22]. Diverse proxies are used to measure ecosystem condition, such as species density and the presence/absence of several species in assemblage structures [23]. A notable advantage of using these aquatic biota is the relative simplicity of their capture and sampling [24, 25]. In particular, the sampling of fish assemblages can be performed through electrofishing, a highly common tool, whereas macroinvertebrate sampling is facilitated and simplified by Surber, D-frame dip, and kick nets (**Figure 2**).

Furthermore, recent studies report that the stomach contents of salmonids (i.e., *Oncorhynchus mykiss* and *Salmo trutta*) contain a diversity of invertebrate prey present in the benthos of nonintervened (hereafter termed “native”) basins, thereby reflecting anthropogenic impacts to the basin [26]. Related to this, Fierro et al. [6] reported similarities in stomach contents and prey diversity of the benthos in river sections with land use different than in the basin.

Likewise, similarities have been found between rivers with more local perturbation, such as through the effects of dams [27, 28]. Therefore, the *O. mykiss* diet might represent an effective bioindicator for evaluating environmental disturbances within the entire basin [6].



**Figure 2.** Left: Fish communities sampled using electrofishing. Right: Aquatic macroinvertebrates sampled using a Surber Net (photograph by P. Fierro).

Among the ecological indices commonly used to evaluate river health, three primary groups exist – biotic indices, multivariate methods, and multimetric indices [15, 19]. Of these, multimetric indices are the most recommended since a large quantity of data can be considered and since these indices may also identify the cause(s) of degradation. This information can then be applied to obtain better understandings of ecosystem status [4]. In turn, biotic indices evaluate river health based only on organism tolerance to organic pollution. One of the most well-known biotic indices is the Hilsenhoff Biotic Index [29], which has been widely used and adapted around the world (e.g., [30–32]). Continuing, multivariate methods require the use of models that relate physicochemical properties of rivers with observed organisms, which are represented under reference (relatively pristine) conditions. These models then compare the observed organisms with those that were “expected.” This comparative method can ultimately detect potentially degraded areas. The most widely used multivariate index is the River Invertebrate Prediction and Classification System [33], which was first implemented in the UK and then adapted to other countries, including Australia [34]. Finally, multimetric indices capture broad characteristic of community structure and function (metric), thus providing a broader understanding of the events occurring in the river [35]. Multimetric indices are powerful tools for establishing the consequences of human activities. These effects may include a high amount of specific and blurred disturbances (nonpoint pollutant discharge), which encompass impacts arising from agriculture, grazing, deforestation, physical alterations of river or bank habitats, dams, sewage discharges, urban areas, and mining [36, 37]. These indices can be applied in several animal assemblages, plant communities, and ecosystems, including terrestrial, marine, and freshwater environments [35]. Corresponding indices of integrity are frequently performed and applied in fish [38] and macroinvertebrates [39]. A summary that contrasts among the three types of indices is presented in **Table 1**.

	<b>Biotic indices</b>	<b>Multivariate methods</b>	<b>Multimetric indices</b>
Examples	Hilsenhoff Biotic Index. Fish Species Biotic Index	River Invertebrate Prediction and Classification System. Australian River Assessment Scheme	Index of Biotic Integrity. Benthic Index of Biotic Integrity
Advantages	Simple, measure only one disturbance (e.g., organic pollution tolerance)	Model created to predict the species and number of organisms that would be expected to appear in a stream system	Include diverse disturbances. Applicable in several animal/plant groups. Incorporates temporal and spatial scale attributes
Disadvantages	Organisms do not respond to only one disturbance; many more stressors affect distribution in the wild	Created models can be easily changed, making the results uncertain. These methods were developed to find patterns and not establish impact	Limited by sampling technique efficiencies. Seasonal migration of biota influences results. Easy confusions with natural perturbations

**Table 1.** Summary of the characteristics considered with stream health indices (adapted from [4]).

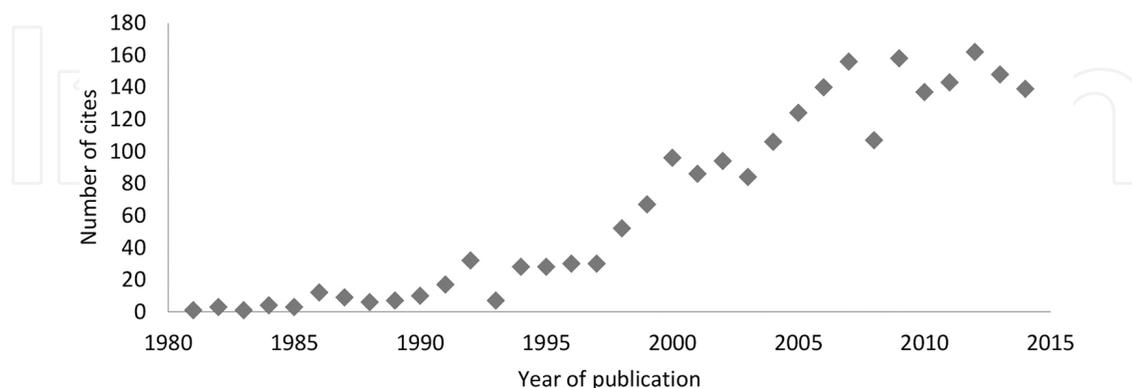
### 3. Assessing the ecological integrity of streams

Ecological integrity, which is also referred to as river health or ecological status, is a measure of the global condition of an aquatic ecosystem. This measurement integrates physical, chemical, and biological integrity elements [15, 17]. Importantly, biological integrity is defined as the ability of aquatic ecosystems to support and maintain a balanced and integrated community with adapted organisms and a composition, diversity, and functional organization comparable to natural habitats within the same region [40–42]. Therefore, a loss of integrity indicates any human-induced positive or negative divergence of the system from a natural, model condition [43].

The Index of Biotic Integrity (IBI), which was initially developed for western USA rivers by [28], is the most used index based on fish assemblages. Consequently, the IBI has been adapted for use to numerous rivers on all continents to evaluate stream health [4, 28]. Indeed, since the creation of the IBI, over 2374 researchers, as of 2014, have used, modified, or mentioned the importance of the IBI (Google Scholar). Furthermore, the number of citations for the IBI grew exponential until 2005, at which point citations “stagnated” near 140 studies per year (**Figure 3**).

Worth highlighting, of the studies presented in this review, the most important milestone occurred from 1986 to 1990. During this period, researchers first began adapting and making modifications to indices based on fish, in addition to these indices being applied in reports to the US government. Between 1991 and 1995, integrity indices were developed for several groups, including macroinvertebrates, birds, and zooplankton. Furthermore, this period was witness to index adaptations to marine and estuary environments. Even terrestrial environments were assessed by the IBI to measure the environmental quality of forests. Between 1996 and 2000, the IBI continued to expand to other groups and environments, such as periphyton

communities, macrophytes, corals, and wetlands. Corresponding adaptations of the IBI to other continents, including Africa, Europe, and South America (Brazil), also occurred [44, 45]. Since 2001, this index is in use on almost all continents and has been adapted several times to different ecoregions within the same countries.



**Figure 3.** Accumulative number of worldwide publications on the index of biotic integrity around the world, starting with the first related publication by [28] (Source: own elaboration).

The advantage of establishing the biotic integrity of rivers based on fish arises as these organisms are present in all, or almost all, rivers, even those that are polluted. Additionally, extensive life history information is available for many species, and fish assemblages generally represent a variety of trophic levels. Indeed, fish are located within the top of the aquatic food chain and can thus help to provide an integrated view of basin environments. Other benefits of the IBI using fish are that fish populations are relatively stable in the summer, when most monitoring occurs; fish are easily identifiable; and the general public can relate to statements about the conditions of fish assemblages. On the other hand, a noted disadvantage of the IBI is that fish are highly mobile, making sampling difficult. Indeed, large groups of personnel, various tools, and an extended period in the field are needed to record daily and seasonal variations [31].

Although less used, the Benthic Index of Biotic Integrity (B-IBI) was developed by Kerans and Karr [46] for rivers of the Tennessee Valley (USA), using the IBI as an initial base [28]. The advantages of using macroinvertebrates as bioindicators are a great biodiversity and an extreme sensitivity and fast response of many taxa to pollution. This quick response is likely due to many macroinvertebrates being sessile and having aquatic life cycles, thus any alterations in environmental limits could lead to death [14]. One significant disadvantage of the B-IBI is that a taxonomic specialist is needed to identify the macroinvertebrate species, which takes a long time. To address this limitation, Rolls et al. [27] used higher levels of taxonomic identification (e.g., genus, family, or both) as a method for adequately describing taxa traits for B-IBI use. Through this technique, a greater cost-benefit might be obtained as less time will be required to taxonomically identify species. Indeed, in countries with few taxonomists and without access to species-level identification keys, application of the B-IBI is very important, as is the case in Chile. Other disadvantages include widespread ignorance about the life histories of many species. Furthermore, it is more difficult for the general public to feel

connected to index results based on macroinvertebrates. Finally, [47] reported that B-IBI requires a large number of samples and multiple metrics to correctly establish the biological condition of a river.

#### 4. Chile: a case study

Mediterranean-climate ecosystems are priority areas of conservation efforts; however, these ecosystems remain threatened globally due to environment degradation [48, 49]. Of the five regions worldwide that present this climate, Chile is the least studied in regards to aquatic ecology [50]. This is despite reporting high national endemism and being considered among the 34 biodiversity hotspots in the world [48, 51].

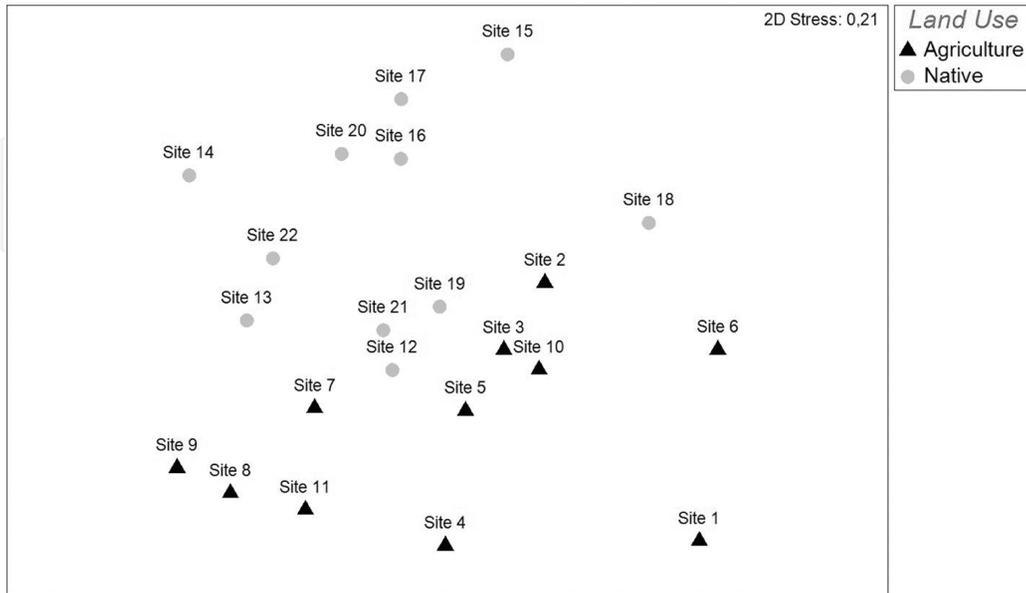
The Mediterranean-climate ecosystem basins of Chile are host to significant industrial activities. This constitutes an increasing problem for aquatic ecosystems due to severe site degradations. Of the various human activities that threaten this region, land use and land cover conversion are highly ranked [52]. Indeed, while many activities directly or indirectly influence aquatic ecosystems, land use is the principal determinant of water quality and of water quantity entering aquatic ecosystems [53]. Furthermore, land cover conversions for crop production or monoculture plantations directly affect freshwater fauna, decreasing, for example, aquatic insect densities and possibly inducing local extinction [9].

In Chile, the use of bioindicators to assess water quality is limited, with applications focused on benthic macroinvertebrate assemblages through a modified Hilsenhoff Biotic Index (e.g., [31, 32, 54]). Notably, these studies were conducted only as a part of basic scientific research as no regulations or laws in Chile stipulate the use of biological criteria for measuring water quality. In contrast, bioindicators are widely used in other countries for assessing and monitoring water quality, often times to meet governmental regulations. In the United States, for example, the Environmental Protection Agency established the “Use of Biological Assessments and Criteria in the Water Quality Program” [55], whereas the European Environment Agency has used biomarker-based monitoring in a number of countries (e.g., Austria 1968 and United Kingdom 1970 [56]).

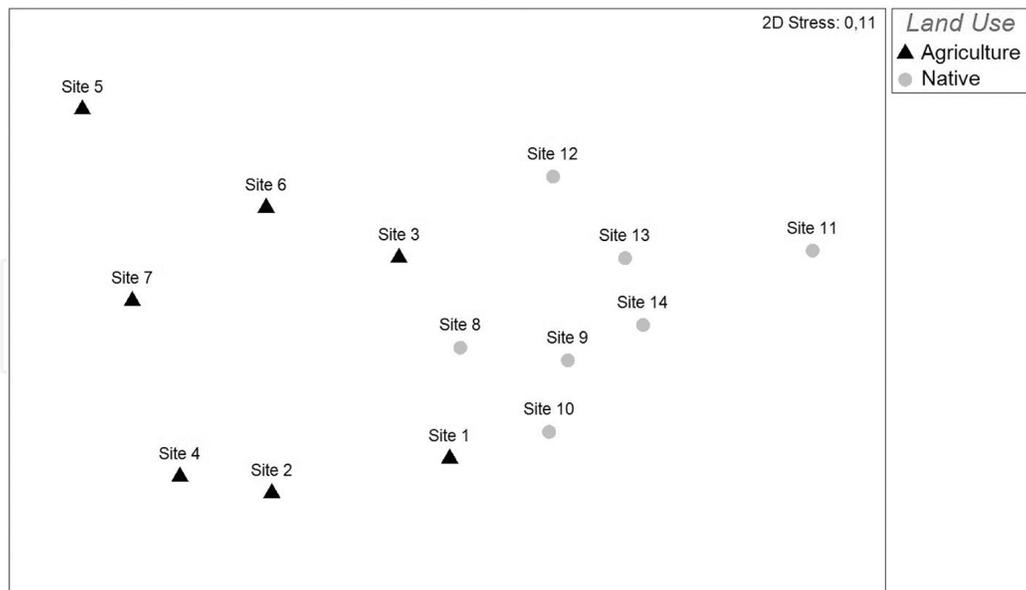
#### 5. Effects of agricultural land use on aquatic ecosystems

Agricultural land use can increase the delivery of several compounds, such as phosphorous and nitrogen, to fluvial ecosystems. In turn, this can produce eutrophication and, consequently, limit the presence of some macroinvertebrate and fish species. For example, when 22 streams were sampled across five Mediterranean-climate watersheds in the farming, central-south region of Chile, agricultural land use was found to be an important predictor of both macroinvertebrate and fish assemblages. Specifically, significant differences in the composition of macroinvertebrate (**Figure 4**; ANOSIM:  $r = 0.203$ ,  $P = 0.01$ ) and fish (**Figure 5**; ANOSIM:  $r = 0.563$ ,  $P = 0.01$ ) assemblages between land use types were found. In addition,

taxonomic diversity of macroinvertebrates was higher in native streams than agricultural streams (average Shannon-Wiener index in native streams: 1.5, agricultural streams: 1.1).

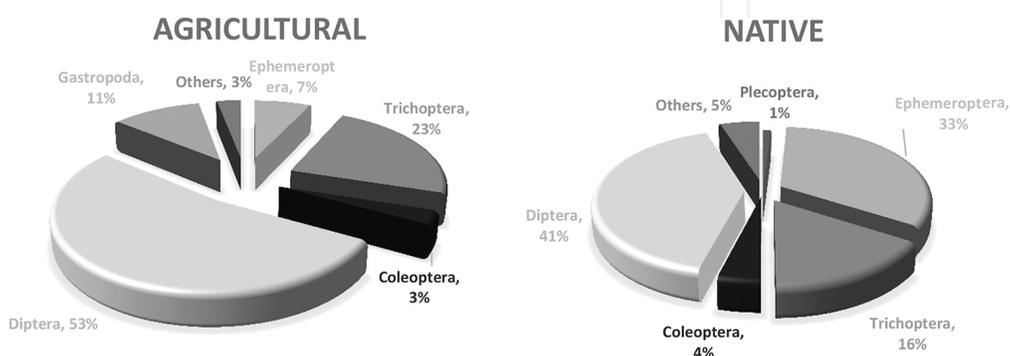


**Figure 4.** nMDS plot based on the composition of macroinvertebrates in 11 native streams and 11 agriculture streams in Mediterranean-climate ecosystems in the farming, central-south region of Chile. The data matrix was constructed using the Bray-Curtis Similarity Index with the square-root transformation of data (9999 restarts). Axes are relative scales and therefore appear without legends (personal data P. Fierro).



**Figure 5.** nMDS plot based on the composition of fish in seven native streams and seven agriculture streams in Mediterranean-climate ecosystems in the farming, central-south region of Chile. The data matrix was constructed using the Bray-Curtis Similarity Index with the square-root transformation of data (9999 restarts). Axes are relative scales and therefore appear without legends (personal data P. Fierro).

The principal difference in both assemblages was community heterogeneity, where native streams were constituted by greater abundances of Ephemeroptera larvae and presented Plecoptera larvae, while in agriculture streams, Diptera larvae and gastropods were more abundant (**Figure 6**). Regarding fish assemblages, a higher amount of taxa were recorded in native streams, and included exotic trout (e.g., *O. mykiss* and *S. trutta*; **Table 2**). These species are unique to environments with low temperatures and high oxygen content, indicators of good water quality. In contrast, the catfish *Trichomycterus areolatus* (**Figure 7**) was recorded at all native and agriculture sites, supporting the broad environmental tolerance of catfish species in general [57].



**Figure 6.** Macroinvertebrate classes found in agricultural dominated and reference streams (N = 22) (unpublished data P. Fierro).

	Agriculture	Native
<i>Diplomystes nahuelbutaensis</i>	0%	4.4%
<i>Trichomycterus areolatus</i>	20.9%	34.1%
<i>Brachygalaxias bullocki</i>	0.2%	0%
<i>Cheirodon galusdae</i>	3.5%	0.6%
<i>Percilia gillisi</i>	20.4%	28.7%
<i>Basilichthys microlepidotus</i>	0%	1.6%
<i>Percichthys trucha</i>	3.2%	0.8%
<i>Gambusia holbrooki</i> *	50.3%	0%
<i>Cnesterodon decemmaculatus</i> *	0.1%	0%
<i>Oncorhynchus mykiss</i> *	1.8%	26.7%
<i>Salmo trutta</i> *	0%	3.2%
<i>Cyprinus carpio</i> *	0.5%	0%

\*Exotic species (unpublished data P. Fierro).

**Table 2.** Species richness and relative abundances of fish species in agriculture and native streams in the farming, central-south region of Chile.



**Figure 7.** Left: Catfish, *Trichomycterus areolatus*, Siluriformes, 9 cm in total length. Center: *Andesiops torrens*, Ephemeroptera, 0.5 cm in total length. Right: *Antarctoperla michaelsoni*, Plecoptera, 0.8 cm in total length. All individuals were collected from streams in the farming, central-south region of Chile (photographs by P. Fierro).

## 6. Conclusion

Macroinvertebrates and fish are used to evaluate the health of streams worldwide. The case results presented in this chapter evidence the importance of using one or more taxonomic groups in bioassessments, where both evaluated assemblages efficiently responded to pressures of human agricultural activities. These results suggest that macroinvertebrates and fish can be used as indicators of water pollution in monitoring programs. Using both assemblages as bioindicators presents several methodological advantages as compared to only assessing physicochemical parameters. These include low costs, easily identifiable fish, and, principally, the sensitivity of both assemblages to different stressors. For example, macroinvertebrates responded differently to substrate compositions than fish, which, in turn, responded to variables such as stream morphometry.

Rivers are increasingly affected by multiple physicochemical and biological stressors. Considering the ongoing rise in environmental management programs for aquatic communities, one related future goal is to develop appropriate indices, such as multimetric or biotic integrity indices, to differentiate between taxonomic groups, thereby facilitating assessments of stream health. However, the effectiveness of these indices will be highly dependent on applicability in different ecoregions.

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